methanol–dichloromethane until there was obtained a filterable solid in a yield of 1.54 g. This material was dissolved in aqueous methanol and treated with decolorizing carbon. The residue obtained on solvent removal was crystallized from methanol–ethanol to yield 1.28 g (72%): mp 195–197°; $[\alpha]^{22}D$ +17 ± 3° (*c* 0.62, methanol); λ_{max}^{KBr} 2.90–3.10 (OH, NH), 6.01, 6.20, 6.40, and 6.80 μ (NH, purine); $\lambda_{max}^{H_2O}$ 262 mμ (ε 14,800); nmr spectrum (deuterium oxide), δ 3.94–5.4 (sugar ring protons and solvent), 6.05 (0.5 proton, distinct doublet, $J_{1,2}$ = 7.9 cps, H-1′), 6.51 (0.5 proton, distinct doublet, $J_{1,2}$ = 6.6 cps, H-1′), 8.22, 8.26, 8.45, and 8.50 (two protons, H-2 and H-8) at 98°, 3.85–4.67 (sugar ring protons and solvent), 6.07 (0.5 proton, distinct doublet, $J_{1,2}$ = 8.0 cps, H-1′), 6.52 (0.5 proton, $J_{1,2}$ = 6.6 cps, H-1′), 8.35, 8.38, 8.46, and 8.54 (two protons, H-2 and H-8); X-ray powder diffraction data 10.65 s (2), 9.50 vw, 7.37 w, 6.19 m, 5.30 s (1), 4.95 m, 4.71 m, 4.13 vw, 3.86 vw, 3.59 s, 3.26 s (3), and 2.90 w.

Anal. Calcd for $C_{10}H_{14}N_6O_3$: C, 45.09; H, 5.30; N, 31.56. Found: C, 45.29; H, 5.55; N, 31.23.

When a solution of the above crystals was examined by thin layer chromatography, with ethyl acetate–methanol (1:1) as developer, there was revealed an elongated spot of R_f 0.30 which gave a positive coloration with ninhydrin. This was later shown (see below) to be two coalescent spots with R_f values of 0.28 and 0.31. Paper chromatography, with 1-butanol–ethanol–water (40:11:19) as developer, revealed a single spot, $R_{adenine}$ 0.62, which was ultraviolet absorbing and gave a positive ninhydrin test.

Separation of the Anomeric Nucleosides.—Following the general technique of Dekker,[7] the above anomeric mixture (700 mg) in methanol–water (1:9) was siphoned onto a 50 × 3.2 cm column of Bio Rad AG 1-X2 (OH^-, 200–400 mesh). Elution was effected with the same solvent and was monitored by an ultraviolet fraction analyzer with 10-ml fractions being collected. At tube 115 an ultraviolet-absorbing component issued from the column. At tube 189 this component was almost completely eluted and a second component appeared. The second component was completely eluted at tube 310. Tubes 115–179 (fraction 1) and 195–310 (fraction 2) were combined separately and evaporated to dryness. The residue from the first fraction was crystallized from water–ethanol to give **9-(2-amino-2-deoxy-α-D-ribofuranosyl)adenine** (**6**) in a yield of 230 mg (33%): mp 149–151°; $[\alpha]^{23}D$ +90 ± 2° (*c* 0.653, methanol); λ_{max}^{KBr} 2.90–3.10 (OH, NH), 6.07, 6.27, 6.42, and 6.82 μ (NH, purine); $\lambda_{max}^{H_2O}$ 262 mμ (ε 14,500); nmr spectrum (deuterium oxide at 98°), δ 3.84–4.60 (sugar ring and solvent), 6.52 (one proton, distinct doublet, $J_{1,2}$ = 6.6 cps, H-1′), 8.38 and 8.54 (two protons, H-2 and H-8); X-ray powder diffraction data 8.84 s (3), 6.60 w, 5.79 vw, 5.48 m, 5.06 s (1), 4.79 w, 4.46 s (3), 4.09 w, 3.73 w, 3.47 m, 3.38 m, 3.23 s (2), 3.01 s, 2.89 w, and 2.86 w.

Anal. Calcd for $C_{10}H_{14}N_6O_3$: C, 45.09; H, 5.30; N, 31.56. Found: C, 45.03; H, 5.15; N, 32.05.

The residue from fraction 2 was crystallized from methanol–ethanol to give **9-(2-amino-2-deoxy-β-D-ribofuranosyl)adenine** (**7**) in a yield of 215 mg (31%): mp 194–196°; $[\alpha]^{22}D$ −66 ± 2° (*c* 0.98, methanol); λ_{max}^{KBr} 2.90–3.10 (OH, NH), 5.88, 6.18, 6.38, and 6.80 μ (NH, purine); $\lambda_{max}^{H_2O}$ 262 mμ (ε 14,400); nmr spectrum (deuterium oxide), δ 3.87–5.14 (sugar ring and solvent), 6.05 (one proton, distinct doublet, $J_{1,2}$ = 7.9 cps, H-1′), 8.22 and 8.45 (two protons, H-2 and H-8); X-ray powder diffraction data 7.97 vw, 7.13 m, 6.65 s (3), 6.02 s (2), 4.90 w, 4.69 m, 4.37 m, 4.21 w, 4.04 w, 3.83 w, 3.63 s (2), 3.40 s (1), 3.18 w, 3.09 vw, 3.01 vw, 2.94 w, 2.85 vw, and 2.77 w.

Anal. Calcd for $C_{10}H_{14}N_6O_3$: C, 45.09; H, 5.30; N, 31.56. Found: C, 45.09; H, 5.11; N, 31.48.

Thin layer chromatography, with ethyl acetate–methanol (1:1) as developer, of the two separate anomers showed two distinct spots with R_f 0.28 (α-D) and 0.31 (β-D). The infrared spectra of the anomers were very similar at all wavelengths except in the region 10.5–12.5 μ. The α-D anomer was less soluble in water than the β-D form.

Registry No.—**4**, 10407-61-1; picrate of **4**, 10407-62-2; **5**, 10407-63-3; **6**, 10407-64-4; **7**, 10414-81-0.

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Synthesis of Guanosine and Its Derivatives from 5-Amino-1-β-D-ribofuranosyl-4-imidazolecarboxamide. I. Ring Closure with Benzoyl Isothiocyanate[1]

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Guanine (VIIa) was synthesized by condensation of 5-amino-4-imidazolecarboxamide (Ia) with benzoyl isothiocyanate followed by methylation and ring closure. This method has also been applied to the synthesis of 2′,3′-O-isopropylideneguanosine (VIIb) from 5-amino-1-(2′,3′-O-isopropylidene-β-D-ribofuranosyl)-4-imidazolecarboxamide (Ib).

5-Amino-1-β-D-ribofuranosyl-4-imidazolecarboxamide (AICA-riboside) is a constituent of 5-amino-1-β-D-ribofuranosyl-4-imidazolecarboxamide 5′-phosphate (AICAR) which has been found to be an important intermediate in the biosynthesis of purine nucleotides.[2,3] Inosine could easily be obtained by treating AICA-riboside with formic acid and acetic anhydride,[4,5] but the synthesis of guanosine has not yet been reported. Since disodium guanosine 5′-phosphate is known to be a useful seasoning agent, development of a new procedure for synthesizing guanosine from AICA-riboside was desirable.

AICA-riboside, the starting material for the present investigation, has been prepared enzymatically[6] or synthetically.[7–9] We used material which was isolated from the culture broth of the mutant of *Bacillus subtilis*[10] and purified by ion-exchange chromatography.

As a preliminary, we investigated the synthesis of guanine (VIIa) from 5-amino-4-imidazolecarboxamide

(1) This paper was presented at the 85th Annual Meeting of the Pharmaceutical Society of Japan, Oct 29, 1965, Tokushima, Japan.

(2) J. M. Buchanan and S. C. Hartman, *Advan. Enzymol.*, **21**, 199 (1959).

(3) S. C. Hartman and J. M. Buchanan, *Ann. Rev. Biochem.*, **28**, 365 (1959).

(4) E. Shaw, *J. Biol. Chem.*, **185**, 439 (1950).

(5) G. Greenberg and E. L. Spilman, *ibid.*, **219**, 411 (1956).

(6) L. N. Lukens and G. N. Buchanan, *J. Am. Chem. Soc.*, **79**, 1511 (1957).

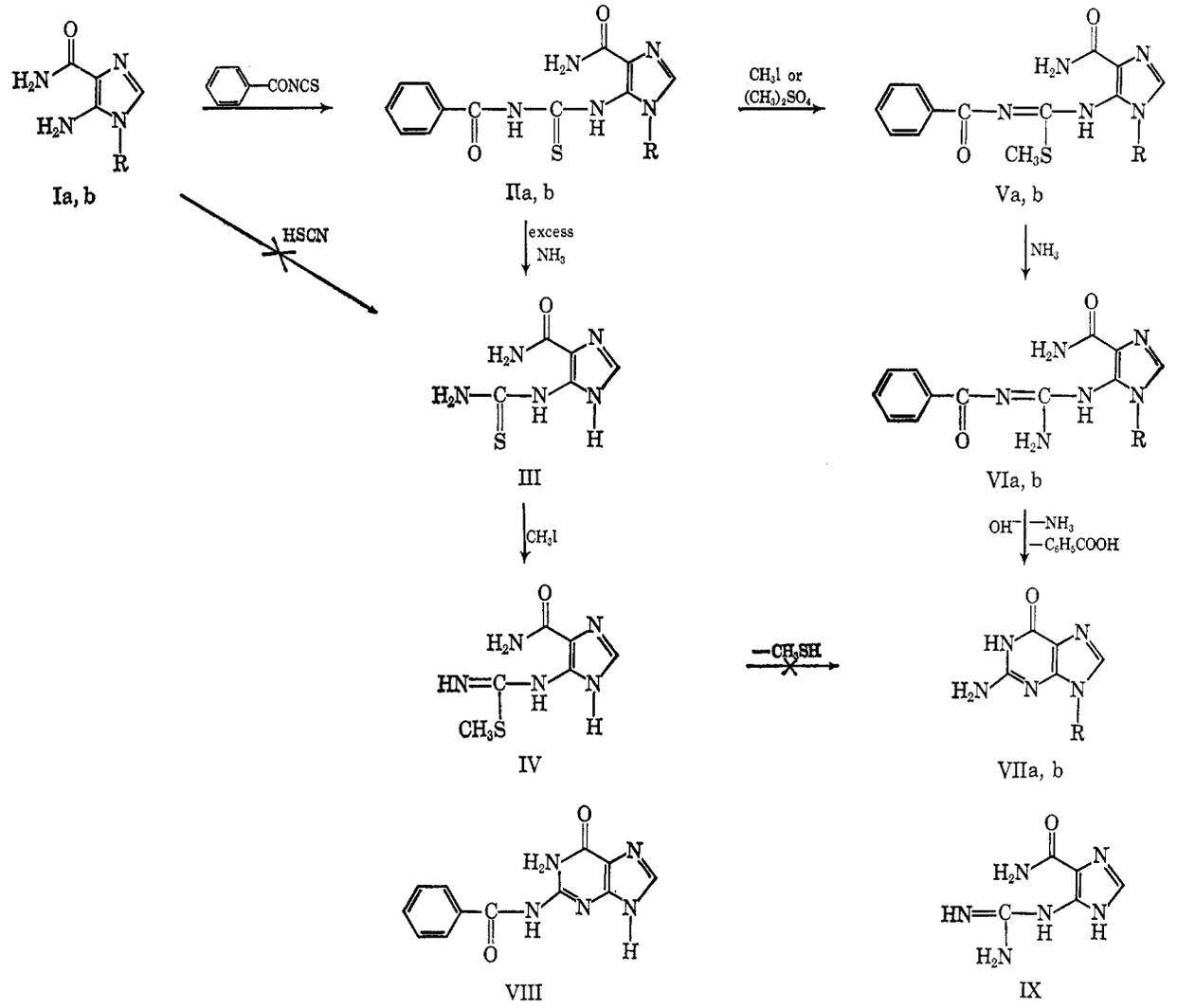
(7) (a) E. Shaw, *ibid.*, **80**, 3899 (1958); (b) *ibid.*, **81**, 6021 (1959).

(8) G. Shaw, R. N. Warrener, D. N. Butler, and R. K. Ralph, *J. Chem. Soc.*, 1648 (1959).

(9) J. Baddiley, J. G. Buchanan, F. E. Hardy, and J. Stewart, *ibid.*, 2893 (1959).

(10) T. Shiro, A. Yamanoi, S. Konishi, S. Okumura, and M. Takahashi, *Agr. Biol. Chem.*, **26**, 785 (1962).

CHART I



a, R = H; b, R = 2′,3′-O-isopropylidene-β-D-ribofuranosyl

(Ia)[11,12] under various mild conditions so as to develop suitable conditions for the synthesis of guanosine (see Chart I). Failure has been recorded in some attempts[9] to prepare VIIa from Ia by means of ring closing agents, such as S-methylisothiourea, cyanamide, and guanidine. Attempts to use several other potential reagents, such as cyanogen bromide, benzoyl cyanamide,[13] and ethyl imidocarbonate,[14] were also unsuccessful. The reaction of Ia with potassium thiocyanate in 10% hydrochloric acid also did not result in the formation of 5-thiocarbamoylamino-4-carbamoylimidazolecarboxamide (III). 5-(N′-Benzoylthiocarbamoyl)amino-4-imidazolecarboxamide (IIa) was, however, readily obtained in 85% yield when Ia was treated with 1 equiv of benzoyl isothiocyanate[15,16] in aqueous solution. However, substitution of acetyl isothiocyanate for benzoyl isothiocyanate did not lead to the corresponding acetylthioureido compound.

(11) E. Shaw and D. W. Woolley, *J. Biol. Chem.*, **181**, 89 (1949).
(12) T. Ichikawa, T. Kato, and T. Takenishi, *J. Heterocyclic Chem.*, **2**, 253 (1965).
(13) A. F. Crowther, *J. Chem. Soc.*, 586 (1948).
(14) J. U. Nef, *Ann.*, **287**, 265 (1895).
(15) A. H. Cook, J. D. Downer, and I. Heilbron, *J. Chem. Soc.*, 1262, 1340 (1948).
(16) A. Takamizawa, K. Hirai, and K. Matsui, *Bull. Chem. Soc.*, **36**, 1214 (1963).

Debenzoylation of IIa with excess ammonia in N,N-dimethylformamide gave III in 85% yield. III was methylated with methyl iodide in alkaline solution to yield 5-(S-methylisothiocarbamoyl)amino-4-imidazolecarboxamide (IV). Although this methylated compound was heated in N,N-dimethylformamide at various temperature, no elimination of methyl mercaptan was detected, and IV was recovered unchanged.

In the hope that the methylmercapto group activated by the electron-attracting tendency of benzoyl group would be more reactive, the methylation of IIa was attempted. Treatment of IIa with a small excess of methyl iodide in aqueous sodium hydroxide solution yielded 5-(N′-benzoyl-S-methylisothiocarbamoyl)amino-4-imidazolecarboxamide (Va) in 77% yield. The resulting methylmercapto derivative was heated in N,N-dimethylformamide at 180° for 5 hr. Contrary to our expectation that the nitrogen atom of the carbamoyl group would attack the carbon atom attached to the methylthio group, N^2-benzoylguanine (VIII) was not obtained.

On heating at 120° for 2 hr in N,N-dimethylformamide containing 2% ammonia in a sealed tube, Va was converted to an imidazole derivative which gave a brown color using Pauly's test. This substance was

easily cyclized to VIIa on treatment with 0.5 *N* sodium hydroxide, ammonia and benzoic acid being eliminated. From these facts as well as from the analytical data, structure VIa was assigned to the product. In this reaction, it seems that VIIa is formed *via* an intermediate which can be either VIII or the guanidino derivative IX. The over-all yield of VIIa based on Ia was about 41%.

AICA-riboside was allowed to react with benzoyl isothiocyanate in water at room temperature, but a benzoylthioureido derivative was not obtained. When the reaction was carried out in 50% aqueous acetone at elevated temperatures (80–100°), a thioureido derivative was detected on paper chromatography in low yield, probably because of partial decomposition of benzoyl isothiocyanate. For the synthesis of guanosine, 5-amino-4-carbamoyl-1-(2′,3′-O-isopropylidene-β-D-ribofuranosyl)imidazole (Ib)[17,18] was employed as a starting material, since Ib is more soluble in organic solvent than AICA-riboside is and since 2′,3′-O-isopropylideneguanosine (VIIb), the ring-closed product to be synthesized, is a direct precursor for 5′-phosphorylation in guanosine 5′-phosphate synthesis. The pK_a values of amino groups for Ia and Ib determined by the potentiometric method were 4.00 and 2.36, respectively.[19] Therefore, the addition reaction of benzoyl isothiocyanate to Ib will require a higher temperature than that for Ia. A mixture of Ib and benzoyl isothiocyanate was then refluxed in acetone, and 5-amino-4-(N′-benzoylthiocarbamoyl)amino-1-(2′,3′-O-isopropylidene-β-D-ribofuranosyl)imidazole (IIb) was obtained in good yield.

IIb was readily methylated with dimethyl sulfate in aqueous sodium hydroxide solution to give the methylthio derivative Vb, which was heated in ethanol containing 2% ammonia to afford the benzoylguanidino derivative VIb. In spite of many attempts, neither IIb, nor Vb, nor VIb could be obtained in crystalline form, but the crude substances showed a single spot on paper chromatography. VIb was heated under reflux in 0.5 *N* sodium hydroxide, neutralized with acetic acid, and evaporated to dryness. Benzoic acid was extracted with hot ethyl ether and the residue was crystallized from water to give VIIb, which was identical with an authentic sample[20] by the specific rotation and ultraviolet and infrared spectra comparison. The over-all yield of VIIb based on Ib determined by paper chromatography and spectrophotometry was 65–70%, and the product was isolated in the pure state in 35–40% yield.

Phosphorylation of VIIb with phosphoryl chloride followed by treatment with aqueous acid gave guanosine 5′-phosphate in good yield.[21]

Experimental Section[22]

Paper Chromatography.—All chromatograms were carried out on Toyo No. 51A filter paper by the ascending technique. Two solvent systems were employed: A, *n*-propyl alcohol—ammonia (28%)—water, 20:12:3 (v/v); B, *n*-butyl alcohol–acetic acid–water, 4:1:1 (v/v).

5-(N′-Benzoylthiocarbamoyl)amino-4-imidazolecarboxamide (IIa).—5-Amino-4-imidazolecarboxamide hydrochloride (Ia) (10 g, 61.7 mmoles) was dissolved in 200 ml of water, and 5.2 g (61.9 mmoles) of sodium hydrogen carbonate was added under stirring. To the neutralized solution was added dropwise 12 g (73.6 mmoles) of benzoyl isothiocyanate, and the mixture was stirred for 1 hr. The resulting precipitate was collected by filtration and crystallized from a mixture of N,N-dimethylformamide and ethanol to afford 15 g (85% yield) of the product: mp 220°; paper chromatography, R_f 0.7 (solvent A) and 0.73 (solvent B); ultraviolet absorption properties, λ_{max}^{EtOH} 244 (ε 14,200), 278 (ε 19,700), and 335 mμ (ε 12,900). This sample showed a band characteristic of thioureido group at 1505 cm^{-1} in the infrared.

Anal. Calcd for $C_{12}H_{11}O_4N_5S$: C, 49.82: H, 3.81; N, 24.22. Found: C, 50.05; H, 4.08; N, 23.92.

5-Thiocarbamoylamino-4-imidazolecarboxamide (III).—IIa (2 g) was added into 20 ml of N,N-dimethylformamide saturated with ammonia at 0° and heated at 100° in a sealed tube for 2 hr. After cooling, the solution was concentrated under reduced pressure and the resulting residue was suspended in 50 ml of hot ethanol with stirring. The insoluble material was filtered and washed with ethanol to yield 950 mg of the crude product. Recrystallization from aqueous N,N-dimethylformamide gave a pure sample: mp 258–260° dec; 850 mg (66%); paper chromatography, R_f 0.45 (solvent A) and 0.35 (solvent B); ultraviolet absorption properties of III, $\lambda_{max}^{pH\ 1}$ 233 (ε 12,600) and 289 mμ (ε 17,300); $\lambda_{max}^{pH\ 13}$ 284 mμ (ε 14,600).

Anal. Calcd for $C_5H_7ON_5S$: C, 32.43; H, 3.78; N, 37.84. Found: C, 32.42; H, 4.37; N, 37.29.

5-(S-Methylisothiocarbamoyl)amino-4-imidazolecarboxamide (IV).—To a stirred solution of III (1.3 g, 6.54 mmoles) in 30 ml of water and 10 ml of 1 *N* sodium hydroxide was added portionwise 1.42 g (10 mmoles) of methyl iodide. The mixture was stirred vigorously for 2 hr at room temperature. The solution was acidified with glacial acetic acid, and the precipitate was collected by filtration. After washing with water, the precipitate was recrystallized from aqueous N,N-dimethylformamide: mp 218–219° dec; 400 mg (28.5%); paper chromatography, R_f 0.52 (solvent B); ultraviolet absorption properties, $\lambda_{max}^{pH\ 1}$ 245 (ε 12,700) and 283 mμ (ε 5700); $\lambda_{max}^{pH\ 13}$ 248 (ε 16,900) and 307 mμ (ε 14,400).

Anal. Calcd for $C_6H_9ON_5S$: C, 36.18; H, 4.52; N, 35.18. Found: C, 35.94; H, 4.80; H, 35.52.

5-(N′-Benzoyl-S-methylisothiocarbamoyl)amino-4-imidazolecarboxamide (Va).—IIa (2 g, 6.92 mmoles) in 0.1 *N* sodium hydroxide (100 ml) was treated with methyl iodide (1.2 g, 8.45 mmoles) at room temperature. After being stirred for 2 hr, the solution was acidified with glacial acetic acid to pH 6. The separated product was recrystallized from glacial acetic acid to give 1.63 g of Va (77%): mp over 250°; ultraviolet absorption properties, λ_{max}^{EtOH} 245 (ε 14,300), 277 (ε 15,000), and 344 mμ (ε 11,800); paper chromatography, R_f 0.72 (solvent A) and 0.70 (solvent B).

Anal. Calcd for $C_{13}H_{13}O_2N_5S$: C, 51.49; H, 4.29; N, 23.10. Found: C, 51.20; H, 4.45; N, 22.59.

5-N′-Benzoylguanidino-4-imidazolecarboxamide (VIa).—Va (2 g) was added to a solution of 50 ml of N,N-dimethylformamide containing 2% ammonia and heated at 120° for 2 hr in a sealed tube. At the end of the reaction the odor of methyl mercaptan was recognized. The solution was evaporated to dryness under reduced pressure, and the resulting residue was crystallized from ethanol to give a crude product (1.4 g). Recrystallization from ethanol gave an analytically pure sample, mp 235°. The crystals were found to be free from sulfur by the nitroprusside test, and Pauly's test[23] gave a brown color. These facts, together with the elementary analysis data, suggest that this compound is the desired VIa: paper chromatography, R_f 0.77 (solvent A) and 0.57 (solvent B); ultraviolet absorption properties, $\lambda_{max}^{Ph\ 1}$ 245 (ε 20,100) and 286 mμ (ε 13,300); $\lambda_{max}^{pH\ 13}$ 257 (ε 20,200) and 320 mμ (ε 11,800).

Anal. Calcd for $C_{12}H_{12}O_2N_6$: C, 52.94; H, 4.41; N, 30.88. Found: C, 52.89; H, 4.80; N, 30.57.

(17) G. Shaw, D. V. Wilson, and C. P. Green, *J. Chem. Soc.*, 2650 (1964).

(18) H. Mori, T. Saito, T. Kato, and T. Takenishi, reported at the Hokkaido Local Meeting of the Chemical Society of Japan, Sapporo, July 1965, abstract, p 11.

(19) Private communication by Dr. T. Meguro in our laboratories.

(20) A. Hampton, *J. Am. Chem. Soc.*, **83**, 3640 (1961).

(21) M. Yoshikawa, K. Kusashio, T. Kato, and T. Takenishi, reported at the 19th Annual Meeting of the Chemical Society of Japan, Tokyo, April 1966, Abstract IV, p 52.

(22) All melting points are uncorrected. Ultraviolet absorption spectra were measured with a Hitachi EPS-2 automatic recording spectrophotometer. Infrared absorption spectra were taken with Jasco Model IR-S spectrophotometer.

(23) B. N. Ames, *J. Am. Chem. Soc.*, **74**, 252 (1952).

Guanine (VIIa).—A suspension of VIa (1.5 g) in 1 *N* sodium hydroxide (40 ml) was refluxed for 3 hr. After neutralization with acetic acid to pH 6, a mixture of benzoic acid and guanine was collected by filtration, washed with water, and dried. The finely divided powder was suspended in 100 ml of hot ethanol with stirring for removing benzoic acid. The purity of the crude guanine (987 mg) was found to be 90% by measuring the ultraviolet absorbancy at 250 mμ. Recrystallization from a minimum amount of 0.5 *N* sulfuric acid yielded the sulfate of VIIa: 540 mg (49%); mp over 300°. This compound was identified with an authentic sample by the comparison of infrared and ultraviolet spectra.

Anal. Calcd for $C_5H_5ON_5 \cdot 0.5H_2SO_4 \cdot H_2O$: C, 27.67; H, 3.93; N, 31.87. Found: C, 27.40; H, 3.65; N, 31.96.

Ia (5 g) was converted to the sulfate of VIIa with an over-all yield of 37–41%. In this case no effort was made to purify the intermediates, and the crude products were directly employed in the subsequent reactions.

2′,3′-O-Isopropylideneguanosine (VIIb) from Ib.—5-Amino-1-(2′,3′-O-isopropylidene-β-D-ribofuranosyl)-4-imidazolecarboxamide (Ib) (6.70 g, 22.48 mmoles) was dissolved in 380 ml of hot anhydrous acetone under stirring. To the solution was added portionwise benzoyl isothiocyanate (4 g, 24.53 mmoles), and the mixture was heated under reflux for 1 hr. During this period, a spot corresponding to the starting material disappeared on paper chromatogram (solvent A) and a new spot appeared. The resulting pale yellow solution was evaporated to dryness *in vacuo*. The yellow powder (11 g) thus obtained was employed in the next reaction: paper chromatography, R_f 0.86 (solvent B). The ultraviolet absorption spectrum of the water extract from an excised spot (R_f 0.86) showed $\lambda_{max}^{pH\ 1}$ 250 mμ.

The benzoylthioureido compound IIb was dissolved in 200 ml of 0.2 *N* sodium hydroxide. To this solution was added portionwise dimethyl sulfate (3.21 g, 25.3 mmoles), and the mixture was stirred vigorously for 2 hr at room temperature. A yellow gummy substance separated. After acidification with glacial acetic acid to pH 6, the gummy substance was extracted four times with 100-ml portions of chloroform. The combined extracts were dried over anhydrous sodium sulfate and filtered. The filtrate was evaporated *in vacuo* to afford amorphous solid (9.5 g.): paper chromatography, R_f 0.93 (solvent B). The ultraviolet absorption spectrum of the extract from an excised spot (R_f 0.93) showed $\lambda_{max}^{pH\ 1}$ 250 mμ.

The S-methylthio derivative Vb, thus obtained, was dissolved in 120 ml of ethanol containing 2% ammonia and heated at 120° for 1 hr in a sealed tube. At the end of the reaction, the odor of methyl mercaptan was recognized. The solvent was removed *in vacuo* to afford a yellow powder (8.9 g): paper chromatography, R_f 0.76 (solvent B). The ultraviolet absorption spectra of the extract from an excised spot (R_f 0.76) showed $\lambda_{max}^{pH\ 1}$ 245 and $\lambda_{max}^{pH\ 13}$ 262 mμ.

Crude N′-benzoylguanidino compound (VIb) was added to 200 ml of 0.5 *N* sodium hydroxide and the solution was heated under reflux for 1 hr. Paper chromatography of this reaction mixture indicated the presence of three spots, 2′,3′-O-isopropylideneguanosine (VIIb), benzoic acid, and a trace of starting material. After neutralization with glacial acetic acid to pH 7, the solution was concentrated *in vacuo* to about 20 ml and the resulting mixture of VIIb and benzoic acid was collected, dried, and pulverized. After the benzoic acid was extracted three times with 200-ml portions of hot ethyl ether, the residue was treated with charcoal and crystallized from water to afford 2.8 g of pale yellow crystals. Recrystallization of the crude substance from water gave an analytically pure sample. The average isolated yield of VIIb was found to be about 35–40%. The pure sample showed mp 290–295° dec; $[\alpha]^{23.8}D$ −67.7° (*c* 1, 0.1 *N* sodium hydroxide). The compound was identified with an authentic sample by comparison of ultraviolet and infrared absorption spectra and R_f values in paper chromatography.

Anal. Calcd for $C_{13}H_{17}O_5N_5$: C, 47.88; H, 5.60; N, 21.52. Found: C, 48.30; H, 5.26; N, 21.67.

When VIIb was synthesized from Ib by the series of reactions described above without isolation of the intermediates, the yield estimated paper chromatographically was 60–65%.

Registry No.—IIa, 10333-86-5; III, 10333-87-6; IV, 10333-88-7; Va, 10333-89-8; VIa, 10333-90-1; sulfate of VIIa, 10333-92-3; VIIb, 362-76-5; benzoyl isothiocyanate, 532-55-8; 5-amino-1-β-D-ribofuranosyl-4-imidazolecarboxamide, 2627-69-2; guanosine, 85-30-3.

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The Crystal and Molecular Structure of the Alkaloid 7-Hydroxy-β-isosparteine Perchlorate

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The structure of an hydroxy-β-isosparteine perchlorate salt has been determined from three-dimensional data collected by an automated diffractometer. The structure was solved by straightforward heavy atom methods and refined by least squares to give unequivocal positions for all of the structural atoms and most of the hydrogen atoms. The hydroxyl group was found to be on the C-7 position, and the proton from the perchloric acid was found on the N-16 position.

The structure of hydroxy-β-isosparteine (Figure 1) is of interest because of its place in the biosynthesis scheme of the *Spartium scoparium* Lupine family of *Leguminosae*, because of the similarity of pharmacological activity of this class of compound to the hormone oxytocin, and because of the unusual p*K* values of the two tertiary nitrogen atoms. The p*K* values of the two nitrogen atoms have been determined to be 2.61 and 11.26 in a similar compound, α-isosparteine.[1] In addition we were interested to see whether the perchlorate group could be used conveniently as a heavy atom group for the determination of a crystal structure.

(1) P. Baranowski, J. Skolik, and M. Wiewioroski, *Tetrahedron*, **20**, 2383 (1964).

Crystal Data

Small needle crystals of the perchlorate salt were given to us by Dr. Marvin Carmack, Department of Chemistry, Indiana University, Bloomington, Ind. The space group and unit cell parameters were obtained from rotation and Weissenberg photographs using Cu Kα radiation. The crystals are orthorhombic with space group $P2_12_12_1$. The unit cell dimensions, with the estimated error in the last figure, are a = 13.04